

WHAT IS CLAIMED IS:

- 1 1. An isolated CLN2 protein with the following characteristics:
 - 2 a) said CLN2 is a protein with pepstatin-insensitive carboxyl protease
 - 3 activity; and,
 - 4 b) mutation or absence of said CLN2 is causative of classical late
 - 5 infantile neuronal ceroid lipofuscinosis (LINCL).
- 1 2. A chimeric protein comprising the CLN2 protein of Claim 1.
- 2
- 3 3. A purified nucleic acid encoding CLN2, or a fragment thereof having at
- 4 least 15 nucleotides.
- 1 4. The nucleic acid of Claim 3 which encodes CLN2 having an amino acid
- 2 sequence as depicted in Figure 3 (SEQ ID NO:3).
- 1 5. The nucleic acid of Claim 4 having a nucleotide sequence as depicted in
- 2 Figure 3 (SEQ ID NO:1), corresponding allelic genes, homologous genes from
- 3 other species, and nucleotide sequences comprising all or portions of *CLN2* genes
- 4 which are altered by the substitution of different codons that encode the same amino
- 5 acid residue within the amino acid sequence (SEQ ID NO:3), thus producing a
- 6 silent change.
- 1 6. The purified nucleic acid of Claim 3 which is DNA.
- 1 7. A recombinant DNA expression vector comprising the DNA of Claim 6,
- 2 wherein the DNA encoding the CLN2 is operatively associated with an expression
- 3 control sequence.
- 1 8. A transformed host cell comprising the DNA vector of Claim 7.

- 1 9. A recombinant virus comprising the DNA vector of Claim 7.
- 1 10. The recombinant virus of Claim 9 selected from the group consisting of a
2 retrovirus, herpes simplex virus (HSV), papillomavirus, Epstein Barr virus (EBV),
3 adenovirus, and adeno-associated virus (AAV).
- 1 11. A method for producing a CLN2 comprising culturing the transformed host
2 cell of Claim 8 under conditions that provide for expression of the CLN2.
- 1 12. The method according to Claim 11 wherein the host cell is a bacterium.
- 1 13. The method according to Claim 11 wherein the host cell is a mammalian
2 cell.
- 1 14. A method for increasing the level of expression of a CLN2 comprising
2 introducing an expression vector of Claim 7 into a host *in vivo* under conditions that
3 provide for expression of the CLN2.
- 1 15. The method according to Claim 14 wherein the expression vector is a viral
2 expression vector.
- 1 16. The method according to Claim 14 wherein the expression vector is a naked
2 DNA expression vector.
- 1 17. A method for treating LINCL in an animal by increasing the level of CLN2
2 in cells.
- 1 18. The method according to Claim 17, wherein the level of CLN2 is increased
2 by administration of CLN2 to the animal.

- 1 19. The method according to Claim 18, wherein the level of CLN2 is increased
2 by administration of a recombinant expression vector to the affected cells, which
3 expression vector provides for expression of the CLN2 *in vivo*.
- 1 20. An oligonucleotide of greater than 20 nucleotides which hybridizes under
2 stringent conditions to the nucleic acid of Claim 3, wherein the T_m is greater than
3 60°C.
- 1 21. The oligonucleotide of Claim 20 which is an anti-sense oligonucleotide.
- 1 22. An antibody specific for CLN2 of Claim 1.
- 1 23. The antibody of Claim 22 which is labeled.
- 1 24. A method for detecting CLN2 in a biological sample comprising:
2 a) contacting a biological sample with an antibody of Claim 22 under
3 conditions that allow for antibody binding to antigen; and,
4 b) detecting formation of reaction complexes comprising the antibody
5 and CLN2 in the sample;
6 wherein detection of formation of reaction complexes indicates the presence of
7 CLN2 in the sample.
- 1 25. A method for quantitating the level of CLN2 in a biological sample
2 comprising:
3 a) detecting the formation of reaction complexes in a biological sample
4 according to the method of Claim 24; and,
5 b) evaluating the amount of reaction complexes formed;
6 wherein the amount of reaction complexes corresponds to the level of CLN2 in the
7 biological sample.

1 26. A method for measuring CLN2 activity in a biological sample based on the
2 amount of CLN2 pepstatin-insensitive carboxyl protease activity relative to normal
3 controls in a biological sample.

1 27. A method for detecting CLN2 in a biological sample comprising:
2 a) contacting a biological sample with an oligonucleotide of Claim 21
3 under conditions that allow for hybridization with mRNA; and,
4 b) detecting hybridization of the oligonucleotide to mRNA in the
5 sample;
6 wherein detection of hybridization indicates the presence of CLN2 in the sample.

1 28. A method for quantitating the level of CLN2 in a biological sample
2 comprising evaluating the quantity of oligonucleotide hybridized according to the
3 method of Claim 26, wherein the quantity of oligonucleotide hybridized corresponds
4 to the level of CLN2 in the biological sample.

1 29. A method for detecting the *CLN2* gene, and mutant variants associated with
2 LINCL, in chromosomal samples comprising of:
3 a) contacting a chromosomal sample from, for example, amniotic fluid,
4 with an oligonucleotide of Claim 21, or variants of said oligonucleotide that
5 hybridize to mutant alleles of *CLN2*, under conditions that allow for
6 hybridization; and,
7 b) detecting hybridization of the oligonucleotide to the chromosomes in
8 the sample;
9 wherein detection of hybridization is used a method of prenatal screening for
10 LINCL.

1 30. A method for identification of lysosomal proteins based on the presence of
2 mannose 6-phosphate glycosylation and comprising the following steps:

- 1 a) purifying proteins from a biological sample using an affinity column
- 2 consisting of the mannose 6-phosphate receptor immobilized on a solid
- 3 support;
- 4 b) peptide sequencing of selected purified proteins;
- 5 c) designing nucleic acid probes based on the peptide sequences derived
- 6 in step b; and,
- 7 d) using the probes of step c to isolate and characterize the genes
- 8 encoding the purified lysosomal proteins.